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STUDIES OF SULFUR OXIDATION AND REACTIONEFFECTS

IN ALBERTA SOILS.

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STUDIES OF SULFUR OXIDATION AND REACTION EFFECTS IN ALBERTA SOILS.

INTRODUCTION.

Sulfur is a component of soils and as such is of primary importance in soil and plant economy. It is a constituent of proteins and therefore essential to life in all its diversity. Sulfur exists in air, usually combined with oxygen, in very small amounts as compared to nitrogen or oxygen. Waksman states that, "Sulfur resembles nitrogen in the transformations into which it enters and in the types of microorganisms which causes these transformations. There is in the sulfur cycle apparent duplications of the processes associated with the nitrogen cycle".

The transformations to which sulfur is subjected in the soil may be summarized under four headings: (1) oxidation (2) reduction, (3) synthesis into proteins, and (4) decomposition of proteins and protein derivatives containing sulfur. Microorganisms are generally conceded to be

essential factors, instrumental in causing the changes of sulfur in its various phases, excepting the synthesizing processes. Purely chemical changes, however, are not excluded as contributory factors in the various transformations.

Sulfur may occur distributed throughout the soil to a considerable depth. It is doubtful, however, if the sulfur below the feeding range of plant roots is of much value as a source of plant food. In certain Alberta soils (76,77) it is not uncommon to find that the relatively soluble salts have leached to greater depths, forming sulfate layers at the point of precipitation of the salts. In badly leached surface soils, therefore, the sulfur present is largely in the form of organic matter and to a somewhat lesser extent in complex insoluble mineral compounds.

The soluble salts of sulfur constitute the source of available sulfur for plant growth; hence, the sulfur present in organic matter and in insoluble form, is not directly available as plant food. The insoluble complex inorganic sulfur compoundsof soils are continually being converted into soluble form through the agency of weathering. By the decomposition of organic sulfur compounds, sulfur is also liberated in some simple form, such as hydrogen sulfide. The hydrogen sulfide and other sulfides in solution are slowly oxidized to sulfur, under natural conditions (71). The sulfur thus formed, as well as elementary sulfur when added to soils, undergoes further oxidation, the resulting end product being sulfuric acid. During the decomposition and oxidation processes, some gaseous sulfur compoundsmay find

their way into the air and thus be lost to plants. However, to compensate for this loss (68,14) some sulfur is continually being returned to the soil dissolved in rain water.

The sulfur oxidizing power of soils is by no means a new subject of investigation. It has been studied by many workers among whom are Johnston (32), who worked with Oregon and Mississouri soils; Joffe and McLean (31), and Halversen and Bollen (19), who worked chiefly with Oregon soils.

REVIEW OF LITERATURE.

The oxidation of sulfides to sulfur, and thence to sulfuric acid, is largely a biological process in which the so-called sulfur bacteria are the acting organisms. According to Waksman (71) the sulfur bacteria were first studied extensively by Winogradsky, who isolated them from sea slime and sulfur springs. Up to the present time the sulfur bacteria from these sources have been investigated more thoroughly than any others.

Morphologically the different groups of sulfur bacteria are not related to one another, but belong to a great many different genera. Waksman (71) divides the sulfur bacteria into five groups on a basis of physiological and morphological differences:

(1) Colorless, thread forming bacteria, accumulating sulfur within their cells.

(2) Colorless, non-thread forming bacteria, accumulating sulfur within their cells.

(3) Purple bacteria, accumulating sulfur within their cells.

(4) Bacteria oxidizing sulfur and sulfur compounds, but accumulating sulfur outside their cells.

(5) Bacteria oxidizing elementary sulfur and not accumulating sulfur within or without the cells.

Members of genus Beggiatoa, belonging to the first group, were the first organisms to attract attention in connection with oxidation of sulfur compounds. In 1870, Cramer pointed out that the minute granules found within the cells of Beggiatoa consisted of sulfur.

Of the five groups mentioned, only the last two members are concerned with the oxidation of sulfur in soils, the other groups, up to the present, have been isolated only from sea waters, fresh waters, sea slime, and sulfur springs.

Nathanson and Beijerinck (1903,1904) were among the first to study members of group four. Nathanson isolated <u>Thiobacillus thioparus</u> from sea water, while Beijerinck discovered <u>Thiobacillus denitrificans</u>. These two organisms or closely related strains, have been isolated from soils. Both bacteria grow best in slightly alkaline media. <u>T. thioparus</u> is stricly aerobic, while <u>T. denitrificans</u> can utilize nitrates as source of oxygen, thus liberating free nitrogen. Both bacteria are autotrophic, that is, development is normal in the absence of organic compounds in the media, the sole source of carbon for metabolic

processes being dissolved carbon dioxide. Demolon (11) found certain ammonifying bacteria capable of oxidizing sulfur to sulfates in inoculated sand and in pure culture media. Joffe (28) working with fungi in pure cultures, supplied with sulfur and insoluble phosphates, found that 40 per cent of the phosphates added went into solution after six months, the reaction not going below pH 4.2. He concluded therefrom that fungi are able to oxidize sulfur.

Only one species, Thiobacillus thiooxidans (65,66), has been definately classified under group five, although there are undoubtedly other forms not yet isolated. This bacterium is aerobic and autotrophic in nature and possesses the extraordinary faculty of flourishing in extremely acid solutions, exhibiting like the nitrate formers a marked indifference to the high concentration of its end product. The optimum reaction of the media is pH 2.0 to 3.0. It is extremely sensitive to the nitrate anion but can withstand concentrations of sulfate as high as 20 per cent of $MgSO_4$. This organism is usually not present in common cultivated soils unless these have been treated with application of sulfur. This bacillus has been found to be the most active in converting insoluble rock phosphate to the soluble form, through the production of sulfuric acid from elementary sulfur, in the Lipman soil-sulfur-rock phosphate compost process (27,44).

Waksman (74) states that the soil reaction, expressed as hydrogen-ion concentration has greater significance in soil biochemical processes than the quantity of acid substance present. In quoting Wollenweber, Waksman states that potato scab is caused by a number of species of Actinomyces, and that these organisms are very snnsitive to acid (.05 per cent) and much less sensitive to alkali (.25 per cent). Waksman, working with pure cultures, found the limiting reaction to vary with the strains of A. scabies, but for nearly all strains the limit was pH 5.0 to 5.2, with pH 4.8 the absolute limit for the most acid resistant forms studied.

The control of the potato scab organism, by regulation of the soil reaction, has not given uniformly satisfactory results, chiefly because of the chemical and physiological complexity of the soil, such as that buffer effect, adsorption power, texture and moisture content; and partly because of the inherent biological characteristics of the potato itself and the diversity of the microflora existing in the various soils.

Sanford (58,59) states that development of the pathogenicity of A. scabies is largely independent of soil reaction between pH 5.4 to 8.5 when dealing with infected tubers. The high acidity limit of groth of the pathogen is in accord with the findings of Waksman (74).

Martin (40,41),on the other hand, obtained partial control with sulfur applications giving a reaction somewhat nearer the point of neutrality and, as a consequence, holds

that regulation of soil acidity is the most promising method of controling A. scabies.

The effect of high hydrogen-ion concentration on the solubility of aluminum and consequent alleged toxicity to plants has been investigated by Magistad and others (39, 38, 67). The results thus obtained indicate that aluminum does not exist in appreciable quantities in solution in soils between pH 4.5 to 9.0.

Increased solubility of calcium and potassium, as a result of sulfur oxidation, is reported by many investigators (1, 61, 46, 47, 67). The extent of solubility of these two constituents appears to vary with different soils depending on the chemical composition.

Other experiments, usually with composts, have been conducted with the intention of converting insoluble phosphates and potassium into soluble form (27, 29, 30, 34, 44, 52, 55). This biological process shows some promise of becoming of practical value, The insoluble phosphatic and potassium rocks do not liberate their constituents in soluble form to any appreciable extent until the reaction reaches pH 2.0 to 3.0 and, consequently, direct application of these constituents mixed with sulfur to soil without previous composting would seem inadvisable.

The addition of sulfur along with the rock phosphate fertilizer treatments of soil has not appreciably increased the availability of the phosphate, due to the

fact that the calcium of the silicate complex is attacked as readily as the calcium of the insoluble phosphate (3).

The use of sulfur and its oxidation product in the reclamation of black alkali soils, by converting the carbonate salts into less harmful compounds, has been the subject of considerable research (21,23,24,51,57). This method of reclamation, in all probability, will only be of value in disposing of residual salts which can only be removed with difficulty by drainage and leaching. The sulfuric acid causes flocculation of the colloidal particles and, hence, considerably aids percolation and leaching of dispersed soils.

Treatments of sulfur, serving chiefly as the source of a limiting element in plant nutrition, has given surprising increases in crop yields, especially of alfalfa, in certain basaltic regions of Oregon and Washington (31,48). Johnston (32) states, as a result of his investigations, that the arid Western soils are low in their sulfate recuperative powers, especially under prolonged cropping, and hence require a larger initial supply of sulfur than do the more humid soils. The greater recuperative power, which is chiefly the result of biological oxidation of the sulfur compounds, explains why a need for sulfur fertilization has not been more apparent in the more humid middle Western soils with a low sulfur and sulfate content.

Again, the question of the effect of sulfur oxidation on ammonification and nitrification has been the subject of

much investigation (2,7,16,37,48). The results obtained indicate that, excluding the heavy applications of sulfur, decomposition of organic matter is not retarded, and that in all soils, excepting slightly buffered acid soils, nitrification is not restricted to any marked extent.

Studies on the relationship of sulfur oxidation to the microflora of the soil (12,54) indicate that small applications of sulfur are somewhat beneficial in increasing bacterial numbers, but larger treatments of sulfur quite definitely reduce the bacterial numbers. The effect of the sulfuric acid apparently is one of partial sterilization of the media.

DESCRIPTION OF SOILS USED IN THE INVESTIGATION.

In the study of the oxidizing power of Alberta soils, samples representing the three major soil belts of the Province were used. A brief description of these soil belts may be of some value in interpreting the results obtained and is therefore presented herewith.

Beaver Lodge Loam. This soil represents the Black Soil Belt, which has its greatest expanse in South-central Alberta and contains approximately seven million acres; much of this acreage is now under cultivation. The soils of this area are normally covered with vegetation consisting of long grass , dotted here and there with mixed willow and polar groves. Over a period of time this type of vegetion has fostered the accoumulation of much residual organic matter and, as a consequence the soils of this area are generally very fertile.

The average annual precipitation varies from 15 to 18 inches, with the heaviest rainfall occurring during the growing months. A favorable moisture distribution together with an absence of warm dry winds may be considered the primary cause of the presence of the Black Soil Belt flora and the consequent accumulation of plant residues.

The Beaver Lodge loam used in this experiment is a virgin black loam, rich in all esential plant food elements and of a high organic matter content. It carries 0.626 per cent of nitrogen and approximately 0.065 per cent of sulfur.

Pigeon Lake Loam: This soil represents the Wooded Soil Belt, which is an area in extent somewhat less than two-thirds of the Province, and occupyihg the northern and north-western parts of the Province. At the present time very little of this soil belt is under cultivation. The soils of this region are usually low in organic matter content, though generally covered with a layer of organic residues up to two or three inches in thickness. This residues consists principally of leaf mold and wood in the various stages of decay. The rapid and complete decomposition of this surface material and the absence of grass roots, percludes the accumulation of much stable residual organic matter, except under swamp conditions. In this belt are found our main muskeg and swamp areas. The annual precipitation is approximately the same as

for the Black Soil Belt. The vegetative covering is largely poplar and spruce and very little grass. This form of vegetation does not foster organic matter accumulation, but, on the other hand, promotes the loss of essential plant food elements from the surface horizon through leaching and hence, owing to the loss of soluble salts, these soils are usually slightly acid in reaction. Being low in organic matter content and depleted in essential elements by leaching, these soils readily respond to corrective treatments of limestone, phosphorus and nitrogen. The absence of residual organic matter and loss of certain salts is reflected in these soils by the grey to light brown color, low water-holding capacity, and poor physical condition. From the standpoint of fertility they are our poorest soils.

The sample used in this study was somewhat higher than the average soil of this area in nitrogen content, as it contained 0.230 per cent of this element. The sulfur content of similar soils is approximately 0.030 per cent.

Brooks Loam: This soil represents the Brown Soil Belt. This belt covers the entire south-eastern part of the Province, consisting wholly of plains land, and covering approximately thirteen million acres. Within this region are found our main "alkali" areas. The soils of this belt are fairly well supplied with essential plant food elements excepting nitrogen. They are, however, considerably lower in organic matter content than the

Black Soil Belt soils; this difference is attributable to a lower average rainfall, which varies from 12 to 15 inches, and the prevalence of warm winds. These two factors combined, have restricted the growth of vegetation and likewise the accumulation of organic matter residue. These factors, also, have prevented the profitable cultivation of much of this area and hence only a small proportion is being cultivated. The brown loam used in this investigation has a nitrogen content of 0.131 per cent; soils from the same locality carry 0.045 per cent of sulfur.

EXPERIMENTAL METHODS.

The organic matter used in the experiment was cured, green sweet clover, finely ground. The superphosphate used is a recent product of the Trail, B.C. smelters; this fertilizer was finely ground to insure thorough distribution within the soil samples. Calcium carbonate (C.P.) was used as a source of lime, and ordinary flowers of sulfur as the elemental sulfur constituent. All experiments were carried on in duplicate.

The soils used in the tumbler experiments were ground to pass a 20 mesh screen and thoroughly air dried at room temperature.100 grams samples of the air-dry soil were weighed into glass tumblers and the computed fertilizer mixtures thoroughly mixed with the dry soil. The tumblers were labelled and distilled water added to the samples to 60 per cent of the water-holding capacity of the soil. The tumblers were stoppered with perforated covers and

then incubated in a dark cupboard at room temperature, which varied from 18 to 25°C. Twice a week the tumblers were stirred with a spatula and more distilled water added, if necessary, to bring the tumblers up to weight.

At the end of the incubation period the samples were weighed and each divided into halves. 25 grams of one half of the moist sample was used for the determination of the hydrogen-ion concentration of the soil sample. The other half of the sample, representing 50 grams of the original sample, was oven dried at 110 to 115°C for 48 hours, after which it was analysed for its water soluble sulfates. The soluble sulfates were determined in the usual way adopted in sulfate analysis, the sulfur being precipitated and weighed as $BaSO_4$. The samples were heated and weighed alternately until two consecutive weighings of each sample agreed within 0.0005 grams of each other.

The soil reaction was determined colorimetrically, using a set of buffer solutions prepared according to Clark (4) and checked against a potentiometer. The moist 25 gram samples, taken as stated above, were placed in shaker bottles and 50 cc. of CO_2-free distilled water added to each; the bottles were then agitated on a wheel for 30 minutes. The supernatant liquid was filtered through a Buchner filter, previously washed with CO_2-free water. The first 10 to 15 cc portion of each filtrate was discarded after having rinsed the receiving flasks.

To 10 cc portions of the main filtrate of each sample, placed in small test tubes, were added the appropriate indicator and the tubes compared with the known standard.

EXPERIMENTAL RESULTS.

The results of the laboratory experiments with sulfur oxidation in the soils from the three soil belts are reported in tables 1 to 4.

As stated elsewhere, the experiments were conducted in duplicate. From a perusal of the data it will be noted that the results reported are based on the averages of the duplicate samples for each period. While in some instances the duplicate analyses failed to produce close agreement, as is frequently the case with biological investigations, yet, on the whole the differences are well within the probable error of the method. Therefore, in the interests of simplicity and to facilitate a numerical comparison of the results obtained, the individual results of analyses have not been included in the tabulated data.

Table 1 gives the results of sulfur oxidation in the Beaver Lodge loam. The samples receiving no sulfur, changed in soluble sulfates to some extent as the incubation period progressed, though on the whole they remained fairly constant.

In considering the rate or velocity of the reaction as distinct from the total amount of sulfate produced at the end of each period there are apparent differences.

Table I: **Oxidation of Elemental Sulfur to Soluble Sulfates in Black Belt Soil (Beaver Lodge Loam) at Various Periods and Under Various Treatments.**

Results expressed as pounds of sulfur per acre (2,000,000 lbs)

Treatment in pounds per acre	2 Weeks		4 Weeks		6 Weeks		8 Weeks		I0 Weeks	
	S in sulfate	Incr. or decr. S oxid. comp. with S alone	S in sulfates	Incr. or decr. S oxid. comp. with S alone	S in sulfates	Incr. or decr. S oxid. comp. with S alone	S in sulfates	Incr. or decr. S oxid. comp. with S alone	S in sulfates	Incr. or decr. S oxid. comp. with S alone
Sulfur 2,000	423		I536		I678		I473		I453	
No treatment	2I2		33I		239		323		3I9	
Diff. due to S	2II		I205		I439		II50		II34	
Clover 20,000 Sulfur 2,000	4I7		559		I270		I227		I466	
Clover 20,000	I46		I62		259		225		277	
Diff. due to S	27I	60	397	-808	I0II	-428	I002	-I48	II89	55
Superphosphate 4,000 Sulfur 2,000	II89		I350		I852		I73I		2063	
Superphosphate 4,000	592		482		60I		677		6I2	
Diff. due to S	597	386	868	-337	I25I	-I88	I054	-96	I45I	3I7
$CaCO_3$ I0,000 Sulfur 2,000	89I		669		I727		I228		I650	
$CaCO_3$ I0,000	385		484		594		35I		295	
Diff. due to S	506	295	I85	-I020	II33	-306	877	-273	I355	22I

For the sulfur treatments the increments at the end of each period over the period immediately preceding (bottom, lst column each period), after subtracting the sulfates of the untreated samples, were as follows: 211, 994, 234, - 289 and - 16 at the end of 2, 4, 6, 8 and 10 weeks respectively. From a consideration of these figures it at once becomes apparent that the velocity of the reaction had reached a maximum at the end of the 4-week period. The peak of the total sulfate accumulation, however, was not reached till after six weeks, when 70 per cent of the added sulfur or 1439 pounds, had accumulated as sulfate. A drop in total sulfates is noticeable at the 8-and 10-week periods. It is doubtful, however, if this difference is significant. Evidently oxidation was only proceeding slowly, if at all, or the sulfate was perhaps reacting with certain soil constituents to form insoluble compounds which, in turn, would not be included in the soluble sulfate figures.

If the results for the supplemented sulfur treatments, after having subtracted the sulfates due to the supplementary fertilizers, are analysed in the same manner it is noticeable at once that they all had the maximum rate of oxidation during the first six weeks of incubation.

From the sixth to the tenth week of incubation there was a slowing down of the rate of oxidation or the sulfates have been transformed to the insoluble state.

The maximum accumulation of sulfates has occurred, in all three cases, at the end of ten weeks.

In order to determine the effect of the supplementary fertilizers on sulfur oxidation, it is necessary to calculate the sulfate increase or decrease of the supplemented sulfur treatments as compared with sulfur alone. In table 4 is a summarized report of the data contained in the second column of each incubation period of tables 1 to 3. From an inspection of table 4, it is apparent that, after a somewhat beneficial effect during the first two weeks of incubation, all supplements have retarded oxidation till after the 8-week period in the Beaver Lodge loam. At the end of the 10-week period there is a beneficial effect in all cases. The addition of organic matter and calcium carbonate caused considerable retardation in the oxidation process during part of the incubation period. The differences attributed to the supplements are in the same direction for all three treatments and moreover, at the beginning of the period, are of such magnitude as to be beyond the limit of the probable error. Where the differences are within the limits of probable error they are, nevertheless, supporting the same conclusion as is derived from the more significant figures.

In order to ascertain if elementary sulfur would be oxidized in dry soil without the aid of much biological activity, an analysis was made of dry soil, and dry

soil to which sulfur had been added at the same rate as used in the tumbler experiment. The average of duplicate samples, treated with sulfur at the rate of 2,000 pounds per acre, gave 234 and 240 pounds of sulfate sulfur per acre at the end of 2- and 4-weeks incubation periods, respectively. The duplicate untreated dry soil samples gave an average of 329 pounds of sulfate sulfur per acre after a 2-week period of incubation. The difference in the results are not considered significant and as the greater sulfate sulfur content was obtained from the untreated samples, serve only to emphasize that little oxidation of sulfur takes place in soil over a short incubation period without the aid of biological agencies.

In table 2 are presented the data covering the sulfur oxidation in the Pigeon Lake loam. The oxidizing power of this soil, as revealed by the figures, is on the whole much lower than that of the Beaver Lodge loam. The results for each treatment where sulfur was added show a gradual upward trend with smaller fluctuations and only a few larger divergencies of any importance (Fig.1).

The untreated samples and the samples treated with calcium carbonate and with organic matter all remain fairly constant and at about the same level in their sulfate content throughout the period of incubation. The super-

Table 2: Oxidation of Elemental Sulfur to Soluble Sulfates in Wooded Belt Soil(Pigeon Lake Loam) at Various Periods and Under Various Treatments.
Results expressed as pounds of sulfur per acre (2,000,000).

Treatment in pounds per acre	2 Weeks		4 Weeks		6 Weeks		8 Weeks		10 Weeks	
	S in sulfates	Incr.or decr.S oxid. comp. with S alone	S in sulfates	Incr.or decr.S. oxid. comp. with S alone	Sinn sulfate	Incr.or decr.S oxid. comp. with S alone	S in sulfates	Incr.or decr.S oxid. comp. with S alone	S in sulfates	Incr.or decr. S oxid. comp. with S. alone
Sulfur 2000	55I		793		564		I049		755	
No treatment	IOI		II9		I47		I5I		94	
Diff. due to S	450		674		4I7		898		66I	
Clover 20000 Sulfur 2000	345		5I3		82I		824		948	
Clover 20000	I40		II8		I28		I52		I79	
Diff. due to S	205	-245	395	-279	693	276	672	-226	769	I08
Superphosphate 4000 Sulfur 2000	53I		923		770		732		I044	
Superphosphate 4000	4I7		42I		44I		443		434	
Diff. due to S	II4	-336	502	-I72	329	-88	289	-609	6I0	-5I
$CaCO_3$ I0000 Sulfur 2000	725		70I		54I		709		93I	
$CaCO_3$ I0000	II5		I82		I58		I58		I26	
Diff. due to S	6I0	I60	5I9	-I55	383	-34	55I	-347	805	I44

phosphate treated samples also remained nearly constant, but at a higher level of sulfates, as the fertilizer contains some sulfate.

The supplementary treatments (Table 4 and fig.I) have had less effect on the rate of sulfur oxidation in this soil than in the Beaver Lodge soil under similar treatment. With two exceptions ($CaCO_3$ after the 2-weeks and clover after the 6-weeks incubation periods), all supplements have depressed sulfur oxidation till after the 8-weeks period. Clover and calcium carbonate produced a beneficial effect during the 10-weeks incubation period. The superphosphate on the other hand, has had a depressing effect on sulfur oxidation throughout the period of incubation. It is noteworthy, however, that the sulfur and the sulfur supplemented treatments, excepting clover, all fluctuated in a most erratic fashion (see table 2). The organic matter on the other hand, had a stabilizing influence on the sulfate production, showing a steady and gradual increase in soluble sulfates as the incubation period progressed (See fig. 1). This observation is in accord with the results obtained with the Beaver Lodge loam, although in the latter the effect is less marked, which is to be expected with soil already rich in organic matter.

For the Pigeon Lake loam the maximum amount

of sulfur oxidized, obtained with the sulfur treatment after eight weeks of incubation, was less than 50 per cent of the quantity of sulfur added.

The sulfur oxidizing power of the Brooks loam was studied separately after the Beaver Lodge and Pigeon Lake experiment had been completed and the data were available. As these data indicated that considerable sulfur remained unoxidized, it was considered desirable to extend the incubation period somewhat in order to ascertain if a greater percentage of sulfur would be converted to sulfates. And, as certain preliminary soil reaction studies had revealed that the reaction was but little affected by the 2,000 pounds sulfur application, a larger application of sulfur (6,000 lbs.) was also included in the experiment. Further, it was decided to limit the scope of the experiment by omitting the 2- and 4- weeks incubation periods. The results of the modified experiment are recorded in table 3.

The Brooks samples receiving no sulfur, like those of the two preceding soils, remained fairly constant throughout in their sulfate content. The superphosphate treatments again were at a higher level of sulfates than the other treatments not receiving sulfur. As the superphosphate undoubtedly contained some sulfates, the results for this fertilizer alone cannot in any sense be considered as indicating the production of sulfates over that of the

Table 3. Oxidation of Elemental Sulfur to Soluble Sulfates in Brown Belt Soil (Brooks Loam) at Various Periods and Under Various Treatments.

Results expressed as pounds of sulfur per acre (2,000,000 lbs.)

Treatment in ounds per acre.	6 Weeks		10 Weeks		14 Weeks.	
	S. in Sulfates	Incr.or decr.S. oxid. comp. with S. alone	S. in Sulfates	Incr.or decr.S. oxid. comp. with S. alone	S. in Sulfates	Incr.or decr. S. oxid. comp. with S. alone
lfur 6,000	1921		2333		3303	
lfur 2,000	1154		1089		1390	
.Treatment	141		163		156	
ff. due to S.	1013		926		1234	
over 20,000 lfur 2,000	958		1270		1305	
over 20,000	88		145		167	
ff. due to S.	870	-143	1125	199	1138	-96
perphosphate 4,000 lfur 2,000	1452		1516		1462	
perphosphate 4,000	511		587		448	
ff. due to S.	941	- 72	929	3	1014	-220
CO_3 10,000 lfur 2,000	1218		1467		1112	
CO_3 10,000	116		198		119	
ff. due to S.	1102	89	1269	343	993	-241

untreated samples. If the sulfates of the untreated soils are subtracted from the superphosphate treated samples after each incubation, for all three soils, it is seen that the figures approach a constant which, with one exception (Beaver Lodge, 4-weeks period) lies between 300 and 400 pounds of sulfate sulfur for the particular rate of superphosphate application (4,000 lbs. per acre).

Where sulfur was applied alone the samples receiving the 2,000 lbs. application had 50 per cent of the sulfur oxidized at the end of 6 weeks and only slightly over 60 per cent at the end of 14 weeks. With the application of 6,000 lbs. of sulfur per acre approximately 30, 37 and 53 per cent of the sulfur had been oxidized at the end of 6, 10 and 14 weeks respectively.

These figures indicate that the sulfur oxidizing microorganisms are incapable of oxidizing as great a percentage of the sulfur when large applications are made, although the total amount of sulfur oxidized is about twice as great with the 3-ton as with the 1-ton application.(upper part table 3).

The sulfur supplements, after some fluctuation throughout the period, appear finally to have had a slightly detrimental effect. The sulfur alone behaved as erratically as any treatment, and it is difficult to draw a definite conclusion as to the effect of the

supplements.

If a comparison of the three soils is made it is apparent, judging from the untreated lots, that the Beaver Lodge loam is from one and one-half to two-times as high in native sulfates (210 to 330 lbs. sulfur) as the two other soils. The Brooks loam is slightly higher in sulfates than the Pigeon Lake loam, (140 to 160 and 90 to 150 lbs. sulfur, respectively). Similarly, if the oxidizing powers of these soils are compared (See fig. 1), they again range themselves in descending order of native sulfates, the Beaver Lodge loam being first and the Pigeon Lake last. The Beaver Lodge soil, however, is only slightly more effective than the Brooks loam,and both are considerably more efficient than the Pigeon Lake soil.

It is quite apparent that all three soils possess a definite and vigorous sulfur oxidizing power capable of converting added sulfur into soluble sulfates.

Again, the effect of the supplements has varied with each soil. It is noticeable that in the Beaver Lodge loam, clover and calcium carbonate caused considerable retardation of the sulfur oxidizing processes. With the Pigeon Lake loam the general tendency was for supplement treatments to cause a retardation of sulfur oxidation, superphosphate the only treatment causing retardation of any importance.

Table 4: The Effect Of Supplements on the Rate of Oxidation of Sulfur, as Compared to Sulfur Alone. (Summary of Second Column, Tables I,2 and 3).

Results expressed as increase or decrease of amount of sulfur oxidized as compared with sulfur alone. (Pounds per acre).

Treatment in pounds per acre	Beaver Lodge Loam					Pigeon Lake Loam					Brooks Loam		
	Weeks Of Incubation												
	2	4	6	8	I0	2	4	6	8	I0	6	I0	I4
Clover 20,000 Sulfur 2,000	60	-808	-428	-I48	55	-245	-279	276	-226	I08	-I43	I99	-96
Superphosphate, 4000 Sulfur 2,000	386	-337	-I88	-96	3I7	-336	-I72	-88	-609	-5I	- 72	3	-220
$CaCO_3$ I0,000 Sulfur 2,000	295	-I020	-306	-273	22I	I60	-I55	-34	-347	I44	89	343	-24I

Fig. 1. Sulfur Oxidized to Sulfates after Varying Incubation Periods. (Tables 1,2 and 3)

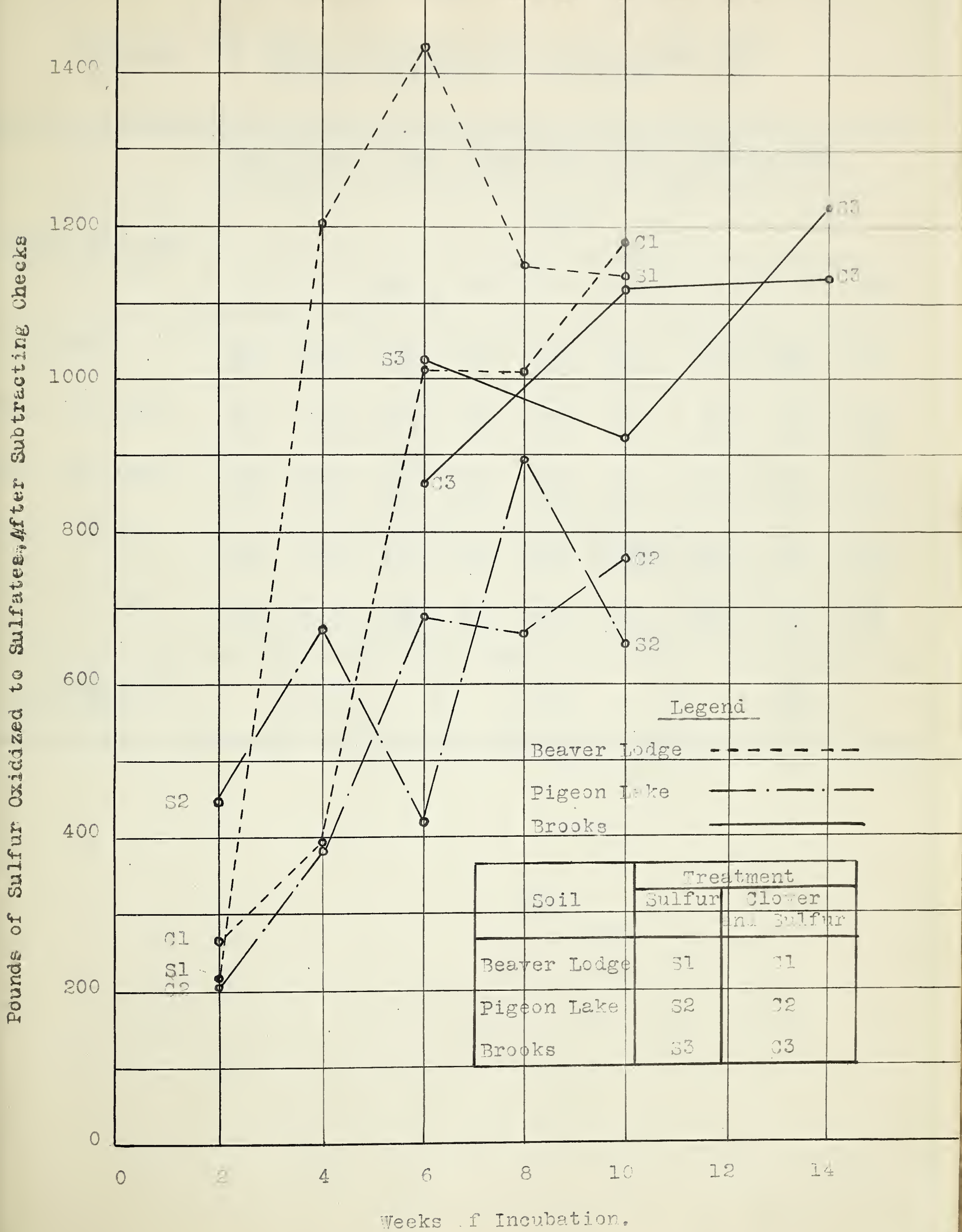

Table 5. The Effect of Various Treatments on the Soil Reaction Expressed as pH Values.

Treatment per acre (2,000,000 lbs.)	Beaverlodge Loam			Pigeon Lake Loam			Brooks Loam		
	Weeks								
	6	10	14	6	10	14	6	10	14
No Treatment	6.2 6.2	6.5 6.5	7.0 7.0	6.0 6.0	5.6 6.0	6.7 6.6	7.0 7.0	7.1 7.2	7.2 7.1
Clover - 10 tons	6.4 6.4	6.4 6.2	6.6 6.6	6.1 6.0	6.2 6.2	6.7 6.6	6.8 6.8	7.0 ---	7.2 7.0
Clover - 20 tons	6.6 6.6	6.5 6.4	6.7 6.7	5.8 6.2	6.4 5.8	6.7 6.7	7.1 7.2	7.1 6.6	7.2 7.2
Sulphur - 1 ton	6.4 6.2	6.5 6.4	6.6 6.6	5.6 5.6	6.4 6.4	6.5 6.4	5.8 5.8	6.4 6.4	6.6 6.6
Sulphur - 3 tons	6.2 6.4	6.4 6.4	6.5 6.5	3.5 4.4	3.4 3.4	3.2 3.2	5.3 5.3	4.2 4.3	4.2 4.2
Soil at beginning of Experiment		6.4 6.4			6.2 6.2			7.2 7.2	

THE EFFECT OF FERTILIZERS ON THE SOIL REACTION.

In table 5 are recorded the results of the investigation into the effect of fertilizers on the soil reaction. It will be noted that certain fertilizers used in the sulfate experiments have been excluded. However, a preliminary part of the investigation had revealed that the excluded fertilizers had no apparent effect on the reaction of the soils under investigation; hence the elimination.

At the beginning of the experiment the air-dry Beaver Lodge and Pigeon Lake soils were slightly acid and the Brooks soil slightly alkaline in reaction. These reactions, however, are all well within the limits for normal productive soils.

The Beaver Lodge loam has remained practically unaffected by all treatments, although there has been a tendency to approach nearer to the point of neutrality as the incubation period progressed. This soil apparently possesses a tremendous buffer capacity as shown by the unchanged reaction after the addition of 6,000 lbs. of sulfur per acre, of which in all probability at least one-half had been oxidized at the end of 14 weeks (Table 3).

For the Pigeon Lake loam the reaction also tended towards neutrality except where sulfur had been applied. The one ton sulfur application had changed the reaction from pH 6.2 to pH 5.6 at the end of 6 weeks. However,

this increase in hydrogen-ion concentration was not maintained at the end of 10 weeks. With the three ton sulfur application there was a pronounced change in reaction, the duplicate samples giving a pH of 3.5 and 4.4 at the end of 6 weeks. This was subsequently reduced to pH 3.4 and 3.2 after 10 and 14 weeks respectively. This soil possesses a lower oxidizing power than the Brooks loam, hence it is safe to assume that less than one-half of the sulfur added had been oxidized at the end of 14 weeks, and yet, if compared with the latter, a higher hydrogen-ion concentration had been obtained. The net result would therefore indicate that the Pigeon Lake loam possesses a somewhat lower buffer capacity than the Brooks loam.

The results for the Brooks loam are very similar to those of the Pigeon Lake loam, if the fact that a slightly alkaline soil was used to begin with, is considered. The one tom sulfur application was sufficient to reduce the pH from 7.2 to 5.8 at the end of 6 weeks. This reaction, however, was not subsequently retained at the end of 10 and 14 weeks. With the three tons application of sulfur a hydrogen-ion concentration of pH 5.3 had been obtained at the end of 6 weeks. This hydrogen-ion concentration was increased to pH 4.2 at the end of 10 weeks and maintained at 14 weeks.

GREEN HOUSE EXPERIMENT.

An experiment was conducted in the green house with Beaver Lodge and Pigeon Lake soils for the purpose of studying the effect of sulfur, supplemented or alone, upon the growth of inoculated alfalfa. Ordinary one-gallon earthenware pots were used as containers. The pots received the same rate of fertilizer application as used in the tumbler experiment. However, the growth of alfalfa was uneven and gave such irregular yields that the data obtained are deemed valueless and consequently are not presented. The reaction was determined on each pot of soil at the end of the experiment, but the results obtained failed to show any significant differences which might explain the irregular yields of alfalfa.

SUMMARY OF METHODS.

The sulfur oxidizing power of three soils, each from one of the major soil belts of the Province, was determined by laboratory experiments. The soils plus their treatments, were placed in tumblers, water was added to 60 per cent of the water-holding capacity of the soils, and the tumblers incubated at room temperature. The soils were analysed at definite intervals over a period of 10 to 14 weeks.

The treatments consisted of sulfur at the rate of 2,000 and 6,000 lbs. per acre. The effects on sulfur oxidation caused by supplementing the sulfur treatments

with calcium carbonate, superphosphate, and organic matter were ascertained.

The sulfur oxidizing power was estimated by determining the amount of sulfur converted to water-soluble sulfates in a given time. The sulfates were determined gravimetrically, being precipitated by $BaCl_2$ and weighed as $BaSO_4$.

From a consideration of the sulfate data, after having subtracted the native soil sulfates and the sulfates due to supplements, the effect of such supplements on sulfur oxidation was determined.

A separate experiment (necessitated by unsatisfactory results obtained with the Berkfeld filters employed at first) was conducted for the purpose of measuring the effect of the sulfur treatments on the hydrogen-ion concentration of the soils. The soil reaction was determined colorimetrically, using a standard set of indicators prepared according to Clark (4). Pot culture experiments with the same soils were conducted in the green house for the purpose of studying the effect of sulfur, alone or supplemented, upon the growth of inoculated alfalfa. The yields of alfalfa obtained were so irregular that the data obtained were considered valueless.

SUMMARY OF RESULTS.

The Black Belt, Brown Belt and Wooded Belt soils under investigation all possess a definite and vigorous

sulfur oxidizing power.

In all three soils the sulfur oxidized at the greatest rate during the first six weeks of incubation. With 2,000 lbs. of sulfur per acre the amounts converted to water-soluble sulfates after 6 weeks incubation were (max. and min. of all treatments) 50-72, 44-50 and 16-35 per cent and after 10 weeks, 57-73, 46-62 and 30-40 per cent for the Beaver Lodge, Brooks and Pigeon Lake soils respectively.

A marked difference exists in the sulfur oxidizing power of the three soils studied. The Beaver Lodge loam possesses the highest oxidizing capacity, the Brooks loam coming next with a somewhat lower capacity and the Pigeon Lake loam ranking third, with only slightly better than one-half the capacity of Beaver Lodge loam.

A significant difference exists in the percentages of sulfur converted to sulfates with sulfur applications of 2,000 and 6,000 lbs. per acre. In the Brooks loam, the percentages converted to sulfates at the end of 6 and 14 weeks of incubation are 29 and 52 per cent, respectively, for the 6,000 lbs. of sulfur, and 51 and 62 per cent, respectively, for the 2,000 lbs. of sulfur application. The actual amount of sulfur converted to sulfates with the 6,000 lbs. sulfur treatment is about twice as great as with the 2,000 lbs. sulfur treatment.

The supplementary fertilizers did not consistentely increase or decrease the rate of sulfur oxidation in the

soils, except in one case, in which superphosphate was added to Wooded Belt soil (Pigeon Lake loam). In this case the quantities of sulfur oxidized were less at the end of each incubation period than where sulfur alone was added. The organic matter (clover) had a stabilizing effect on sulfur oxidation in all soils tending to produce a steadier increase in sulfur oxidized throughout the periods of incubation.

No direct correlation exists between the amounts of sulfur oxidized and the change in hydrogen-ion concentration with treatments up to 6,000 lbs. of sulfur per acre.

The 2,000 lbs. application of sulfur had produced a significant change at the end of 6 weeks in the Brooks and Pigeon Lake loams. This depression in pH value, however, was not sustained at 10 and 14 weeks, the soils having reverted approximately their original reaction.

With the 6,000 lbs. of sulfur application the Pigeon Lake and Brooks loam experienced approximately the same numerical change in pH values. Using the dry soil pH as basis, the change in the reaction, over the same period, was from pH 7.2 to 4.2 and 4.2. These results tentatively indicate that between 2,000 and 6,000 lbs. of sulfur per acre would be required to inhibit the potato scab organism under laboratory or green house conditions.

The Beaver Lodge loam possesses a tremendous buffer

capacity, as even the larger sulfur applications failed to create any apparent increase in the hydrogen-ion concentration of this soil.

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